Exhibit I

26. Wiskerchen M, Muesing MA.
Human immunodeficiency virus type 1 integrase: effects of mutations on viral ability to integrate, direct viral gene expression from unintegrated viral DNA templates, and sustain viral propagation in primary cells. J Virol. 1995 Jan;69(1):376-86
[PubMed ID: 95074888]

Abstract:
Integrase is the only viral protein necessary for integration of retroviral DNA into chromosomal DNA of the host cell. Biochemical analysis of human immunodeficiency virus type 1 (HIV-1) integrase with purified protein and synthetic DNA substrates has revealed extensive information regarding the mechanism of action of the enzyme, as well as identification of critical residues and functional domains. Since in vitro reactions are carried out in the absence of other viral proteins and they analyze strand transfer of only one end of the donor substrate, they do not define completely the process of integration as it occurs during the course of viral infection. In an effort to further understand the role of integrase during viral infection, we initially constructed a panel of 24 HIV-1 mutants with specific alanine substitutions throughout the integrase coding region and analyzed them in a human T-cell line infection. Of these mutant viruses, 12 were capable of sustained viral replication, 11 were replication defective, and 1 was temperature sensitive for viral growth. The replication defective viruses express and correctly process the integrase and Gag proteins. Using this panel of mutants and an additional set of 18 mutant viruses, we identified nine amino acids which, when replaced with alanine, destroy integrase activity. Although none of the replication-defective mutants are able to integrate into the host genome, a subset of them with alterations in the catalytic triad are capable of Tat-mediated transactivation of an indicator gene linked to the viral long terminal repeat promoter. We present evidence that integration of the HIV-1 provirus is essential not only for productive infection of T cells but also for virus passage in both cultured peripheral blood lymphocytes and macrophage cells.